

Invasive and Exotic Species

Reproductive Potential of Triploid Grass and Black Carp

Chinese carps, introduced into waters of the United States, have established self-sustaining populations in many areas. These exotic species have degraded and modified aquatic habitats thereby negatively impacting native fish and wildlife populations. Adverse effects of exotic or invasive species have been observed on both ecological and recreational uses of our aquatic resources. In fact, feral black carp have recently been captured in the Mississippi River.



Triploid and diploid grass carp have been introduced throughout the Mississippi River Basin to control unwanted aquatic vegetation and have since established self-sustaining populations in many areas.

Some states permit the introduction of genetically altered, or triploid, carps under the assumption that these animals are sterile, unable to reproduce. However, there are studies that indicate methods of both induction and detection of triploidy may not be 100% effective. The release of diploid organisms into the environment would result. Additionally, some triploid organisms can produce viable gametes and offspring which may allow triploid fishes to establish reproductively sustainable populations.



Now, triploid black carp are being proposed as a solution to parasitic trematode problems in the aquaculture industry. Black carp feed primarily on mollusks and pose a serious threat to native Mississippi River basin bivalves should they establish populations in the wild.

Investigations on-going at CERC have objectives to 1) compare the accuracy of methods used to verify triploidy; 2) evaluate the reproductive potential of triploid and diploid black and grass carps; and 3) determine population characteristics (growth and survival rates) of black carp.

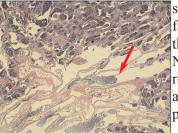
The aquaculture industry standard for identification of triploid carp is currently by the Coulter Counter® method*. We verified the efficacy of this technique against flow cytometric analysis, a more precise method used in research laboratories. Flow cytomtery identified 2 diploid black carp in 1000 presumed triploid fish as tested by the Coulter Counter® method.

'In a Coulter Counter®, cells are passed through electrodes, which measure the resistance in particles size. Triploids have larger blood cells than diploids so more resistance is measured in the triploids. Flow cytometry quantifies DNA content in an individual cell. Since diploid cells contain two sets of chromosomes and triploids contain three sets, triploids have 1.5 times the DNA of diploids.



Milt collection from a diploid grass carp male.

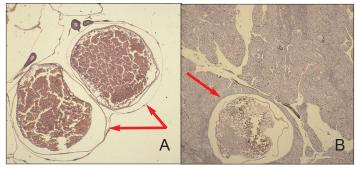
Initial experiments have been conducted to assess reproductive success of the carps. Carps estimated to be 3-6 years old were injected with pituitary hormones to induce



spawning. The female fish failed to release eggs, but the males produced milt. Necropsy and histology revealed that both triploid and diploid grass carp males produced sperm.

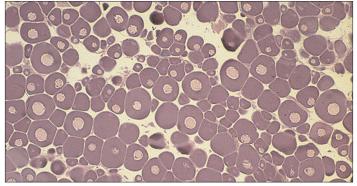
Histological section of testing a triploid grass carp. Sperm production is visible (arrow).

Female diploid grass carp contained ripe eggs but did not ovulate in this study. Triploid grass carp ovaries predominately contained oocytes arrested at early stages of development; however maturing oocytes we identified scattered throughout the organ.



Histological section of ovaries. A) Adult diploid grass carp showing mature atretic oocytes (arrows). B) Triploid grass carp showing mostly undeveloped oogonia with one atretic oocyte (arrow).

Ovaries of female diploid black carp contained immature oocytes and thus were apparently too young to spawn.



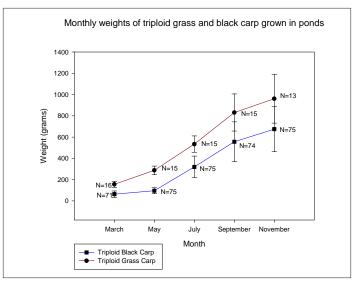
Histological section of diploid black carp ovary showing primary oocytes.

Subsequent studies will attempt to spawn specific intraspecies crosses of diploid and triploid carps, as well as inter-species hybridization of grass x black carp. We hypothesize that milt from triploid male black carp could fertilize eggs of diploid female grass carp.

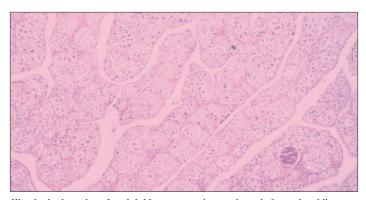
Currently fingerling diploid and triploid grass and black carps, verified by flow cytometric analysis, have been individually pit-tagged and are being reared under secured conditions at CERC to monitor growth and gonadal development.



Fish are individually identified by inserting a PIT tag into the body cavity.



Growth of triploid grass and black carp grown in ponds.



Histological section of a triploid grass carp (approximately 6 months old) showing normal development of ovarian tissues at this stage.

Collectively, these data will allow us to make predictions about the potential for black carp to establish populations in the Mississippi River Basin. The hazards are real and these studies are designed to allow quantitative estimation of the risks posed by these species of Chinese carp.

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